

Chapter 7

The Control of Microbial Growth

The Terminology of Microbial Control

- Sepsis refers to microbial contamination
- Asepsis is the absence of significant contamination
- Aseptic surgery techniques prevent microbial contamination of wounds
- Sterilization: Removing all microbial life
- Commercial sterilization: Killing *C. botulinum* endospores
- Disinfection: Removing pathogens
- Antisepsis: Removing pathogens from living tissue
- Degerming: Removing microbes from a limited area
- Sanitization: Lowering microbial counts on eating utensils
- Biocide/germicide: Kills microbes
- Bacteriostasis: Inhibiting, not killing, microbes

Microbial Death Curve

Effectiveness of Treatment

- Depends on
- Number of microbes
- Environment (organic matter, temperature, biofilms)
- Time of exposure
- Microbial characteristics

Actions of Microbial Control Agents

- Alteration of membrane permeability
- Damage to proteins
- Damage to nucleic acids

Heat

- Thermal death point (TDP): Lowest temperature at which all cells in a culture are killed in 10 min
- Thermal death time (TDT): Time during which all cells in a culture are killed

Decimal Reduction Time (DRT)

- Minutes to kill 90% of a population at a given temperature

Moist Heat Sterilization

- Moist heat denatures proteins
- Autoclave: Steam under pressure

Steam Sterilization

- Steam must contact item's surface

Pasteurization

- Reduces spoilage organisms and pathogens
- Equivalent treatments
- 63°C for 30 min
- High-temperature short-time: 72°C for 15 sec
- Ultra-high-temperature: 140°C for <1 sec
- Thermophilic organisms survive

Dry Heat Sterilization

- Kills by oxidation
- Dry heat
- Flaming
- Incineration
- Hot-air sterilization

Filtration

- HEPA removes microbes >0.3 µm
- Membrane filtration removes microbes >0.22 µm

Physical Methods of Microbial Control

- Low temperature inhibits microbial growth
- Refrigeration
- Deep-freezing
- Lyophilization
- High pressure denatures proteins
- Desiccation prevents metabolism
- Osmotic pressure causes plasmolysis

Radiation

- Ionizing radiation (X rays, gamma rays, electron beams)
- Ionizes water to release OH•
- Damages DNA
- Nonionizing radiation (UV, 260 nm)
- Damages DNA
- Microwaves kill by heat; not especially antimicrobial

Principles of Effective Disinfection

- Concentration of disinfectant
- Organic matter
- pH
- Time

Use-Dilution Test

- Metal rings dipped in test bacteria are dried
- Dried cultures are placed in disinfectant for 10 min at 20°C
- Rings are transferred to culture media to determine whether bacteria survived treatment

Disk-Diffusion Method

Phenol & Phenolics

- Disrupt plasma membranes

Bisphenols

- Hexachlorophene, triclosan
- Disrupt plasma membranes

Biguanides

- Chlorhexidine
- Disrupt plasma membranes

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Halogens

- Iodine
- Tinctures: In aqueous alcohol
- Iodophors: In organic molecules
- Alter protein synthesis and membranes
- Chlorine
- Bleach: Hypochlorous acid (HOCl)
- Chloramine: Chlorine + ammonia
- Oxidizing agents

Alcohols

- Ethanol, isopropanol
- Denature proteins, dissolve lipids
- Require water

Heavy Metals

- Ag, Hg, and Cu
- Silver nitrate may be used to prevent gonorrheal ophthalmia neonatorum
- Silver sulfadiazine used as a topical cream on burns
- Copper sulfate is an algicide
- Oligodynamic action
- Denature proteins

Surface-Active Agents, or Surfactants

Chemical Food Preservatives

- Organic acids
- Inhibit metabolism
- Sorbic acid, benzoic acid, and calcium propionate
- Control molds and bacteria in foods and cosmetics
- Nitrite prevents endospore germination
- Antibiotics
- Nisin and natamycin prevent spoilage of cheese

Aldehydes

- Inactivate proteins by cross-linking with functional groups ($-\text{NH}_2$, $-\text{OH}$, $-\text{COOH}$, $-\text{SH}$)
- Use: Medical equipment
- Glutaraldehyde, formaldehyde, and ortho-phthalaldehyde

Gaseous Sterilants

- Denature proteins
- Use: Heat-sensitive material
- Ethylene oxide

Plasma

- Free radicals destroy microbes
- Use: Tubular instruments

Supercritical Fluids

- CO_2 with gaseous and liquid properties
- Use: Medical implants

Peroxygens

- Oxidizing agents
- Use: Contaminated surfaces
- O_3 , H_2O_2 , peracetic acid

Endospores and Mycobacteria